

Fbx15 Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Fbx15

Project type

Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

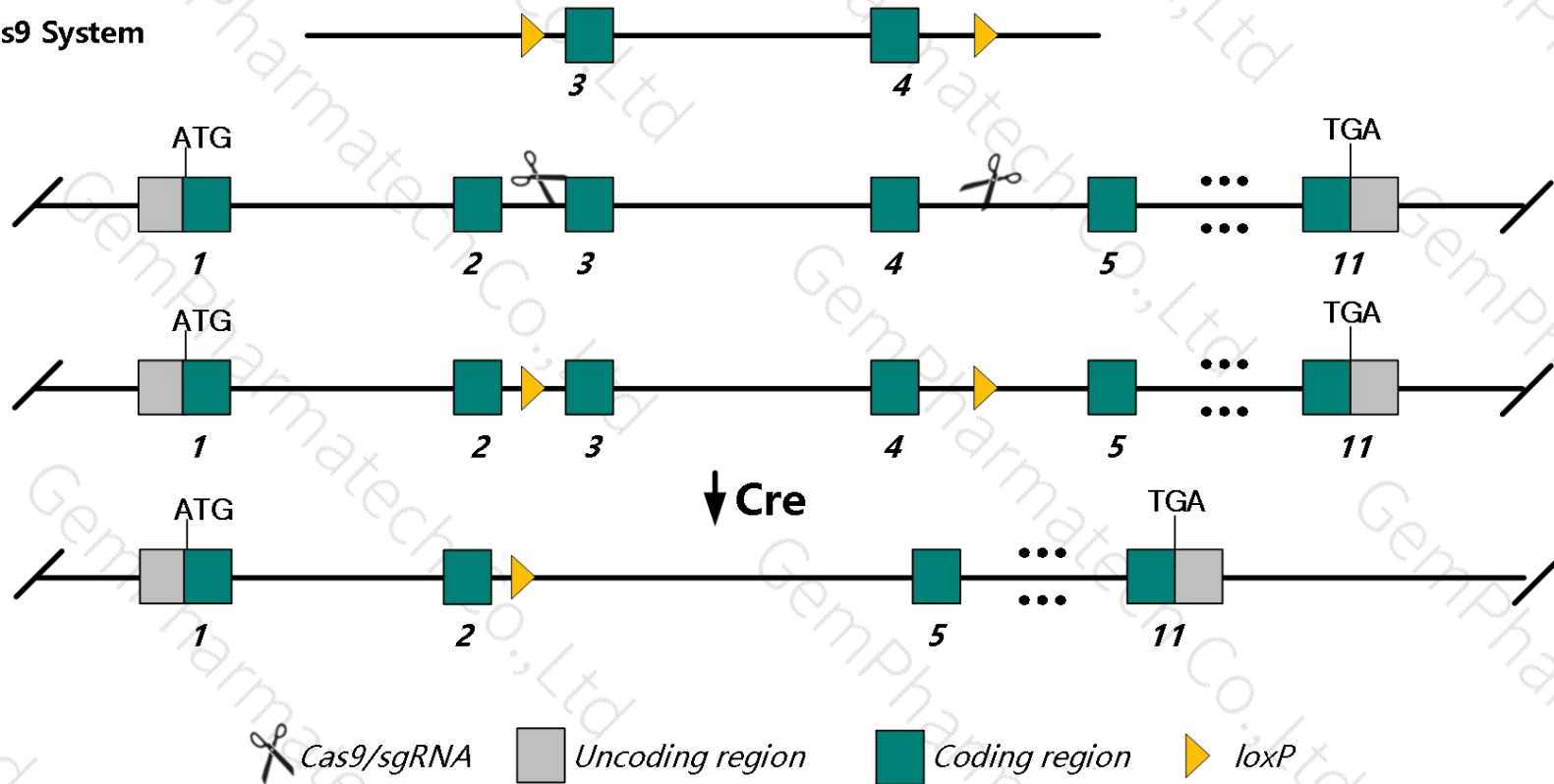
This model will use CRISPR/Cas9 technology to edit the *Fbxl5* gene. The schematic diagram is as follows:

Donor and CRISPR/Cas9 System

Wild-type allele

Floxed allele

KO allele



- The *Fbxl5* gene has 12 transcripts. According to the structure of *Fbxl5* gene, exon3-exon4 of *Fbxl5*-201 (ENSMUST00000047857.15) transcript is recommended as the knockout region. The region contains 283bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Fbxl5* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed. Cas9, gRNA and Donor were microinjected into the fertilized eggs of C57BL/6JGpt mice. Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. A stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6JGpt mice.
- The flox mice will be knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues and cell types.

- According to the existing MGI data, Mice homozygous for a null mutation display embryonic lethality before turning of the embryo with iron overload, growth retardation, and hemorrhage. Mice heterozygous for a knock-out allele exhibit abnormal iron homeostasis when fed a low iron diet.
- The *Fbxl5* gene is located on the Chr5. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.
- This Strategy is designed based on genetic information in existing databases. Due to the complexity of biological processes, all risk of loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information (NCBI)

Fbxl5 F-box and leucine-rich repeat protein 5 [*Mus musculus* (house mouse)]

Gene ID: 242960, updated on 12-Aug-2019

Summary



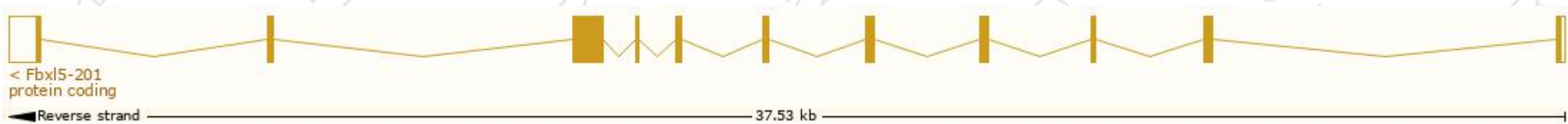
Official Symbol	Fbxl5 provided by MGI
Official Full Name	F-box and leucine-rich repeat protein 5 provided by MGI
Primary source	MGI:MGI:2152883
See related	Ensembl:ENSMUSG00000039753
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Fbl4; Fir4
Expression	Ubiquitous expression in cerebellum adult (RPKM 17.0), placenta adult (RPKM 16.2) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

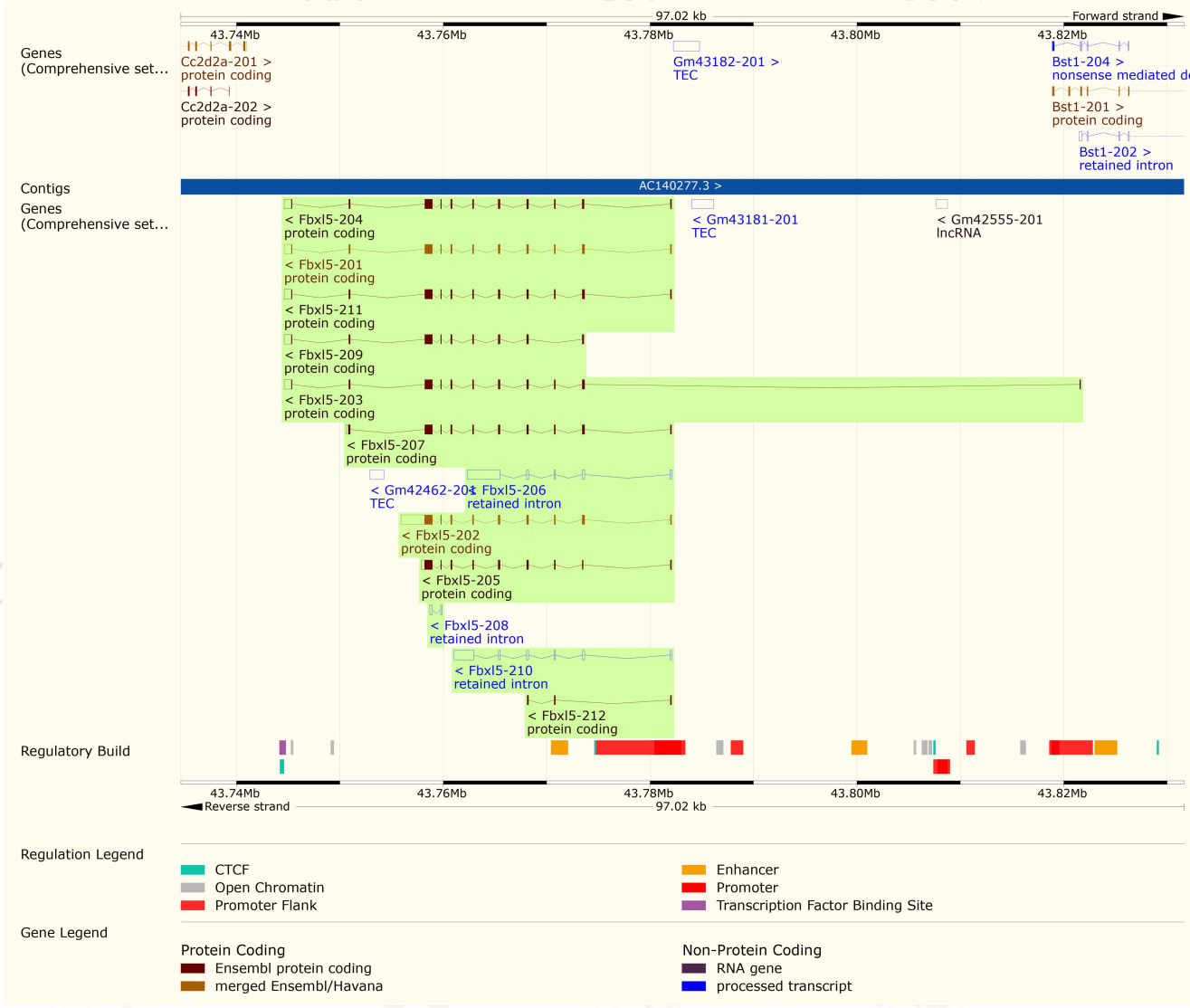
The gene has 12 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Fbxl5-202	ENSMUST00000087465.10	4237	623aa	Protein coding	CCDS19263	Q8C2S5	TSL:1 GENCODE basic
Fbxl5-201	ENSMUST00000047857.15	2858	690aa	Protein coding	CCDS51489	Q8C2S5	TSL:1 GENCODE basic APPRIS P2
Fbxl5-211	ENSMUST00000196483.4	2814	689aa	Protein coding	-	Q8C2S5	TSL:1 GENCODE basic APPRIS ALT1
Fbxl5-204	ENSMUST00000119523.7	2801	673aa	Protein coding	-	D3Z584	TSL:5 GENCODE basic
Fbxl5-203	ENSMUST00000114047.9	2762	684aa	Protein coding	-	Q8C2S5	TSL:1 GENCODE basic
Fbxl5-209	ENSMUST00000141902.7	2517	611aa	Protein coding	-	F6W6I1	CDS 5' incomplete TSL:5
Fbxl5-207	ENSMUST00000124610.5	2306	679aa	Protein coding	-	F7BZC4	TSL:5 GENCODE basic
Fbxl5-205	ENSMUST00000121736.5	2144	580aa	Protein coding	-	Q8C2S5	TSL:1 GENCODE basic
Fbxl5-212	ENSMUST00000199055.1	374	107aa	Protein coding	-	A0A0G2JE06	CDS 3' incomplete TSL:5
Fbxl5-206	ENSMUST00000124421.1	3833	No protein	Retained intron	-	-	TSL:1
Fbxl5-210	ENSMUST00000143316.7	2731	No protein	Retained intron	-	-	TSL:1
Fbxl5-208	ENSMUST00000140469.2	397	No protein	Retained intron	-	-	TSL:2

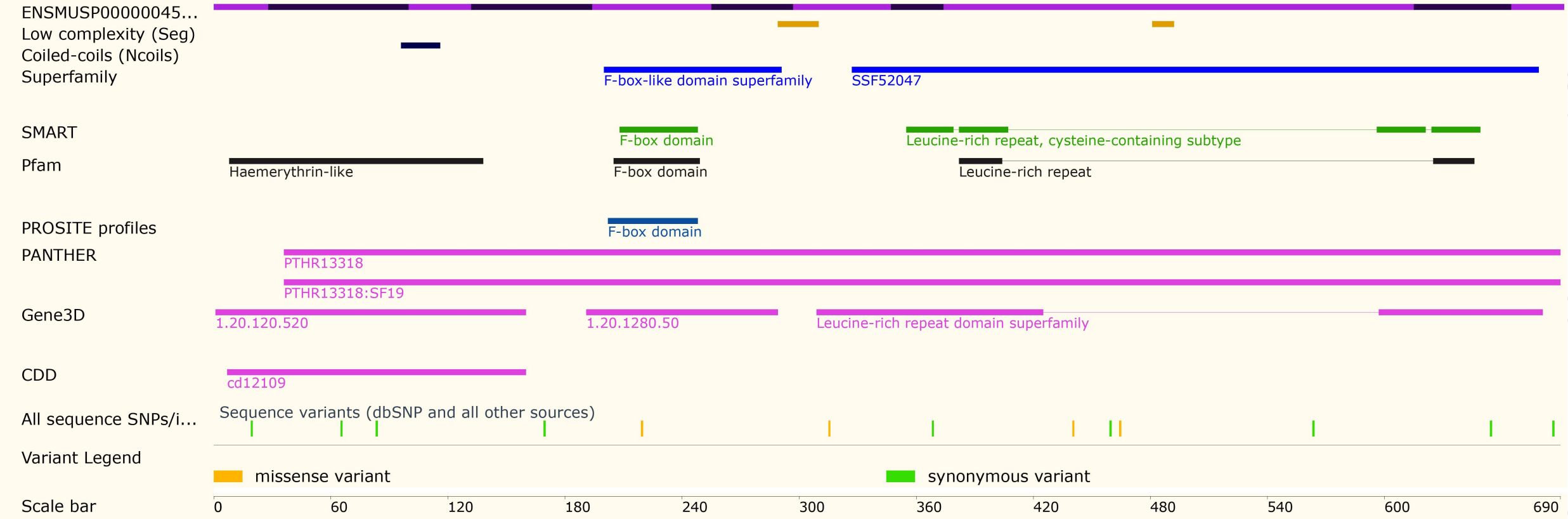
The strategy is based on the design of *Fbxl5-201* transcript,The transcription is shown below



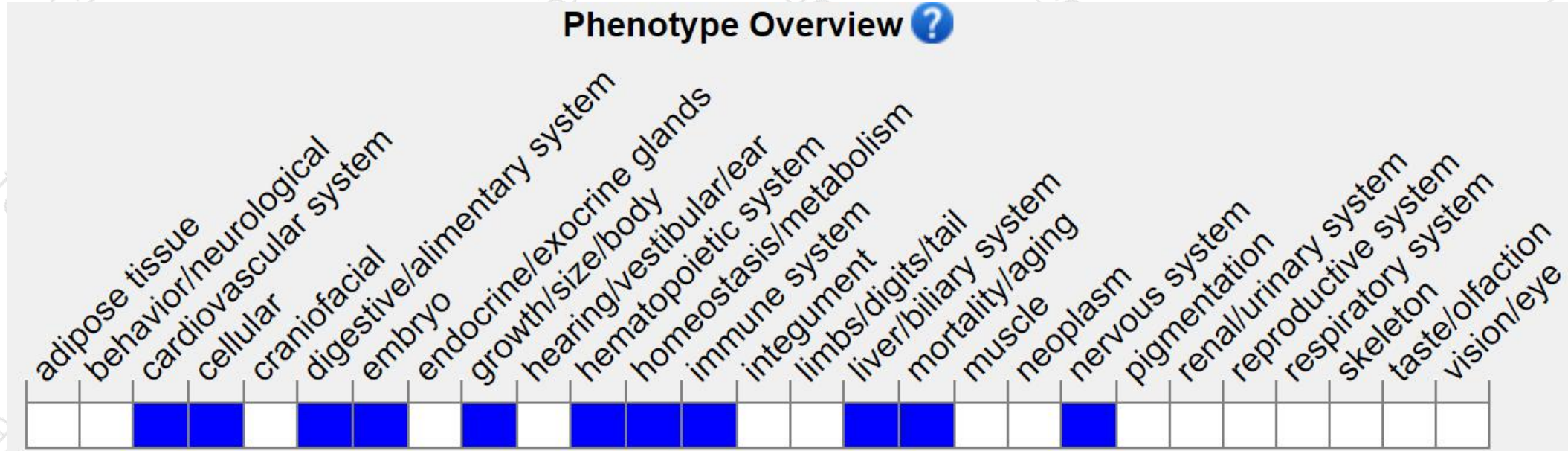
Genomic location distribution



Protein domain



Mouse phenotype description(MGI)



Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(<http://www.informatics.jax.org/>).

According to the existing MGI data, Mice homozygous for a null mutation display embryonic lethality before turning of the embryo with iron overload, growth retardation, and hemorrhage. Mice heterozygous for a knock-out allele exhibit abnormal iron homeostasis when fed a low iron diet.

If you have any questions, you are welcome to inquire.

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